

“Investigation of iron injection impact on iron metabolism biomarkers to detect blood transfusion”

Dr. N. Leuenberger, Dr. B. Favrat (Swiss Laboratory for Doping Analyses, Switzerland)

Project overview

Proteins involved in erythropoiesis and iron metabolism have been demonstrated as potential biomarkers to detect blood doping. Hepcidin, a peptide hormone that is a regulator of iron homeostasis, is reported to be regulated by recombinant human erythropoietin (rhEPO) administration and by the autologous blood transfusion. It was suggested that measurement of iron in ethylenediaminetetraacetic acid (EDTA) plasma may be a cost-effective marker for the screening of blood transfusion use. Increased levels of EDTA plasma iron were also detectable using a fast automated method. Thus, EDTA plasma iron may provide further evidence of blood manipulation.

In 2014, erythroferrone (ERFE) was identified as a novel erythroid regulator of iron metabolism in a mouse model. These observations suggest that ERFE may be a potential biomarker in the detection of blood doping. However, the ironomics strategy to detect blood doping could be impacted by iron injection performed by athletes and be considered as a confounding factor for iron metabolism biomarkers.

In this project a clinical study will be performed to investigate the potential confounding effect of iron injection on iron metabolism biomarkers.

Results and Conclusions:

Proteins involved in iron metabolism, such as hepcidin, are potential blood doping biomarkers. Monitoring of these markers could offer a strategy to complement the actual longitudinal follow-up of hematological parameters measurement, such as reticulocyte percentage (Ret%), to detect blood doping. Iron injections, which are frequently supplied to athletes, could affect hepcidin concentration and therefore, present a confounding factor in hematological profiling for blood doping detection. In this project, urinary iron was tested as a novel biomarker to monitor iron injection.

A randomized, single-blind, placebo-controlled trial was conducted in male volunteers who received a single intravenous injection of ferric carboxymaltose or placebo. The effects of iron injection on iron metabolism markers and hematological were investigated. Hepcidin concentration was measured in blood by liquid chromatography coupled with high-resolution mass spectrometry (LC-HRMS) and the urinary excretion kinetics of iron was quantified by inductively coupled plasma mass spectrometry (ICP-MS).

Intravenous iron supplementation increased Ret% significantly, and also serum hepcidin concentration was increased 16-fold relative to baseline.

Interestingly, urinary iron concentration was increased by 12-fold at 3 h after iron injection, and it remains significantly elevated until day 1 after administration. A specificity of 100% and a sensitivity of 79.3% were achieved with a proposed threshold of 251 ng/mL for urinary iron.

Due to the impact on Ret%, iron injection is a confounding factor in evaluation of the hematological parameters for the detection of blood doping. Furthermore, urinary iron quantification could offer a novel strategy to monitor intravenous iron injection in the doping control samples.